J Neurosurg 55:669–677, 1981

Catecholamine neurotransmitters, psychoactive drugs, and biological clocks

The 1981 Harvey Cushing oration

JULIUS AXELROD, PH.D.

Section on Pharmacology, Laboratory of Clinical Science, National Institute of Mental Health, Bethesda, Maryland

√ In his Cushing oration, the 1970 Nobel Laureate reviews the experimental history of the vital role which chemical agents play in the transmission of nerve impulses and the important functions of the brain. He reveals the intriguing steps in his own early involvement in the field of neurotransmitters. A beacon for neuroscientists of the future is his unique talent for not only looking, but seeing potentially significant clues.

KEY WORDS • **catecholamine** • **neurotransmitter** • **sympathetic nervous system** • **psychoactive drug** • **pineal gland** • **β-adrenergic receptor** • **circadian rhythm**

I would like to talk to you today about my researches on chemical nerve transmission, a field that I worked in from 1957 to about 1977. Chemical neurotransmission has a rather romantic history. The first proposal about chemical neurotransmission was made by a graduate student at the University of Cambridge, T. R. Elliot, in 1905. Elliot noted that when he injected an extract of the adrenal gland in a dog, the organs responded as if the sympathetic nervous system were being stimulated. Just prior to this time, John J. Abel, a pharmacologist at Johns Hopkins University, had isolated the active pressor principal of the adrenal gland and identified it as the catecholamine, adrenaline. Elliot then proceeded to inject adrenaline into the dog and noted again that the organs responded as if the sympathetic nerves were stimulated. He proposed the brilliant concept that nerves exert their effect on target organs by the liberation of a chemical similar to adrenaline. He then presented his idea at a meeting of the British Physiological Society.[8] But his professor, the noted physiologist Langley, disapproved. He told Elliot that unless he could prove that a nerve does release a chemical he should not talk about it; and in Elliot's full paper in the *Journal of Physiology*, he did not mention his ideas on chemical neurotransmission.

This concept struck the imagination of several scientists, particularly Otto Loewi, an Austrian pharmacologist. One evening he had a dream. Loewi dreamt of an experiment to prove chemical transmission of nerve impulses, and he jotted it down on a pad on his night table. When Loewi woke up the next morning, he couldn't read his handwriting. However, very shortly after that, he dreamt the same dream again and he immediately dashed to the laboratory to do the experiment. It was an elegant experiment. What he did was to immerse two frog hearts in a common bath. When he stimulated the vagus nerve of one frog heart, the beat of the second heart was slowed, indicating that the nerve was releasing a chemical that affected the beat of the second heart.[17]

Another pioneer in chemical neurotransmission was Walter Cannon, a professor of physiology at Harvard. He too noted that a chemical, similar to adrenaline, was liberated into the blood stream of dogs in fear and anger. There was great controversy among scientists who believed in electrical transmission and those who believed in chemical transmission. It was around the middle of the 1940's that this controversy was settled. Ulf von Euler, a Swedish physiologist, isolated the active principal of the sympathetic nerve and identified it as noradrenaline.[24] Sir Henry Dale, a few years earlier, identified acetylcholine as the transmitter of the cholinergic nervous system.

By the 1950's, three catecholaminergic neurotransmitters had been identified: adrenaline, noradrenaline, and dopamine. Adrenaline at that time was considered to be a hormone arising from the adrenal medulla. It is released into the blood stream and acts on distant organs. Noradrenaline is a transmitter of sympathetic nerves. This catecholamine is involved in private communications between nerves. Dopamine is the Cinderella of the catecholaminergic transmitters. It was believed for many years that its only function was to serve as a precursor to noradrenaline. However, in the last 7 or 8 years, it has been shown to have important functions in the brain and is concerned in movement, activity, mood, and endocrine function.

My interest in neurotransmission came about quite accidentally. In 1955, I joined the National Institute of Mental Health from the National Heart Institute. My lab chief told me that I could work on anything I pleased, and I started to do research on liver enzymes because I felt comfortable with this type of investigation. At a seminar one day, Seymour Kety, who was the head of our laboratory, reported on a fascinating observation made by two Canadian psychiatrists, Osmond and Hoffer. What they had found was that when adrenaline was allowed to be exposed to the air, it turned pink. And when they injected this "pink" adrenaline into human subjects they hallucinated. Hoffer and his co-workers[12] then proposed that perhaps psychoses may be due to an abnormal metabolism of adrenaline. This struck me as a fascinating concept.

Adrenaline Metabolism Studies

A great deal of my previous work was concerned with the metabolism and distribution of drugs in the body. A drug that I worked on was amphetamine which is very closely related to adrenaline in chemical structure. I thought a study on what happened to adrenaline in the body would be a great problem and would be appropriate for a National Institute of Mental Health scientist. In 1957, hardly anything was known about the metabolism of adrenaline. I spent 3 frustrating months looking for an enzyme that makes "pink" adrenaline. I couldn't find it. And then one day I came across an intriguing abstract by a biochemist, Armstrong. He and McMillan reported that subjects with pheochromocytomas excreted a compound which they called "3-methoxy vanilmandelic (VMA) acid."[1] What struck me about it was it had a methyl group on the oxygen and it surely must be coming from adrenaline or noradrenaline, since these catecholamines are highly concentrated in pheochromocytoma. Then that very afternoon I did a crucial experiment. A page from my notebook will give you an idea about the experiment (Fig. 1). I suspected that the methyl group in VMA should be coming from the methyl donor, S-adenosylmethionine. I did not have this compound, but I knew that S-adenosylmethionine arises from adenosine triphosphate (ATP) and the amino acid, methionine. I made a rat liver preparation and added ATP, methionine, and adrenaline, and the adrenaline disappeared. When either ATP or methionine was omitted, there was little disappearance of adrenaline. This experiment told me that adrenaline was being metabolized by O-methylation and that S-adenosylmethionine arising from ATP and methionine was the methyl donor. I won't go into the boring details, but we found a new metabolite, O-methyl adrenaline, which was named "metanephrine," and a new pathway for the metabolism of catecholamines.[3] As a result of these and other experiments, it was proposed that adrenaline or noradrenaline was metabolized by two pathways: the first by O-methylation via catechol-O-methyltransferase (COMT) which I isolated and characterized, and the other by deamination by monoamine oxidase. The irony of this whole story was that when Kety and his colleagues looked for differences in adrenaline metabolism in patients with schizophrenia, they found none at all.

Inactivation of Neurotransmitters

One important characteristic of a neurotransmitter is that its actions must be rapidly terminated, otherwise they would persist and cause many problems. In 1957, it was thought that the actions of neurotrans-

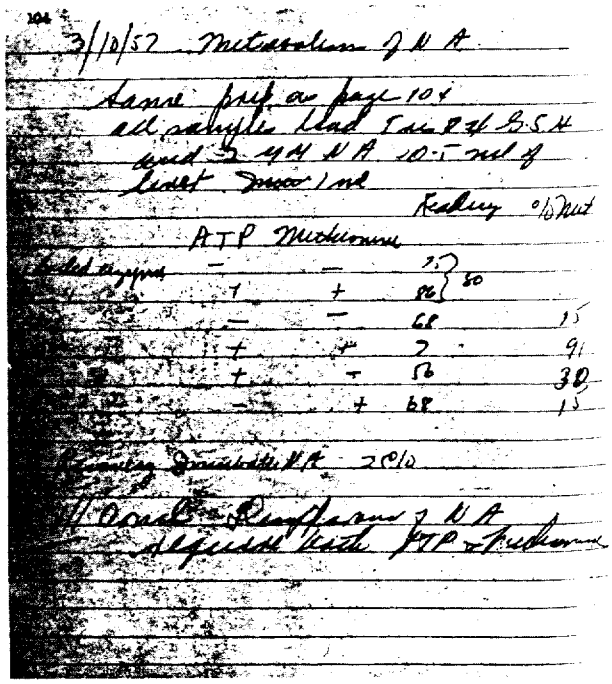

FIG. 1. A page from my notebook of an experiment that led to the discovery of the O-methylation pathway for catecholamines and the enzyme, catechol-O-methyltransferase.

mitters were ended by enzymes. If this were true, then COMT and/or monoamine oxidase would be involved. When these enzymes were almost completely inhibited by drugs and noradrenaline was injected into animals, the pressor action of noradrenaline was still rapidly terminated. This experiment indicated that these enzymes had very little to do with rapidly ending the pressor actions of noradrenaline. What do you do next if a pet hypothesis doesn't hold? Gordon Whitby, a visiting scientist in my laboratory, injected radioactive noradrenaline into cats and examined its distribution in tissues. We observed that ^{3}H-noradrenaline was highly localized in tissues that were rich in sympathetic nerves.[25] This suggested to us that it might be taken up into the nerves. If noradrenaline was taken up into the nerves it could be an effective way to rapidly terminate the actions of this neurotransmitter. But how do you prove it? My colleagues and I exchanged many ideas about experiments to prove that noradrenaline is taken up in the nerves. One day, George Hertting, who was a visiting scientist, and I thought of an experiment to prove uptake of noradrenaline in nerves. It was very simple and I think neurosurgeons will appreciate this experiment. We removed the superior cervical ganglion unilaterally in cats, allowing the nerves to degenerate on one side only, and then we injected ^{3}H-noradrenaline. We found that in the denervated side there was hardly any ^{3}H-noradrenaline present (Table 1).[11] In the innervated side, there were large amounts of the radioactive noradrenaline. This indicated that the neurotransmitter was taken up in the nerves and served as a mechanism for inactivating the neurotransmitter. This concept was unique and could be used to explain many actions of drugs, as I will tell you shortly.

In another experiment, we infused radioactive noradrenaline into the dog gracialis muscle and then we stimulated the nerves to this muscle. There was a marked release of noradrenaline.[18] In still another experiment, we added phenoxybenzamine, an α-adrenergic blocking agent. We found that in the presence of the phenoxybenzamine more noradrenaline was released. We misinterpreted the results. We reported that phenoxybenzamine increased the amount of ^{3}H-noradrenaline release by blocking its uptake. It was not so. Fifteen years later, it was shown by others that the nerves have presynaptic inhibitory α-adrenergic receptors. If these receptors are inhibited by α-adrenergic blocking agents, a greater amount of the neurotransmitter is released. A compelling proof of uptake was to demonstrate the presence of ^{3}H-noradrenaline in nerves after its injection. Therefore, in another experiment, ^{3}H-noradrenaline was injected and the pineal gland, an organ rich in sympathetic nerves, was subjected to autoradiography. Radioactive grains were found to be highly localized in the sympathetic nerves.[26] These nerves were also found to contain dense core granules in which the transmitters were being stored.

TABLE 1

*Effect of denervation on the uptake of ^{3}H-noradrenaline**

Tissue	Chronic Denervation		Acute Denervation	
	Den	Inn	Den	Inn
external ocular muscle	5.9	41.9	24.9	26.6
retractor bulbi muscle	1.9	11.3	13.2	13.4
lacrimal gland	3.2	44.7	—	—
salivary gland	5.7	42.4	75.5	88.6

* Values for ^{3}H-noradrenaline are ng/gm tissue. In the chronic experiments, the right superior cervical ganglia were removed from adult male cats under pentobarbital anesthesia. After 21 days, cats were given ^{3}H-noradrenaline intravenously under anesthesia. One hour later, the cats were decapitated and the ^{3}H-noradrenaline assayed. In the acute experiments, the right superior ganglia from two cats were removed 15 minutes before ^{3}H-noradrenaline was administered. Den = denervated; and Inn = innervated. (Modified from Hertting G, *et al.*: Lack of uptake of catecholamines after chronic denervation of sympathetic nerves. *Nature 189*:66, 1961.)

At just about this time, histological techniques were developed by Swedish investigators to visualize noradrenergic nerves.[9] The tissues were exposed to formaldehyde gas which condensed with the catecholamine neurotransmitter in the nerves and formed an intense fluorophor. The terminals visualized by this technique showed many swellings or varicosity-like beads on a string (Fig. 2). A single cell body has about 25,000 varicosities. These biochemical, pharmacological, and histological observations formed the basis for what has been proposed as a model of the sympathetic neuron[2] (Fig. 2). Within the varicosities, there are dense core vesicles that store the transmitter. Upon stimulation of the nerve, the vesicles fuse with the plasma membrane and then form an opening. The

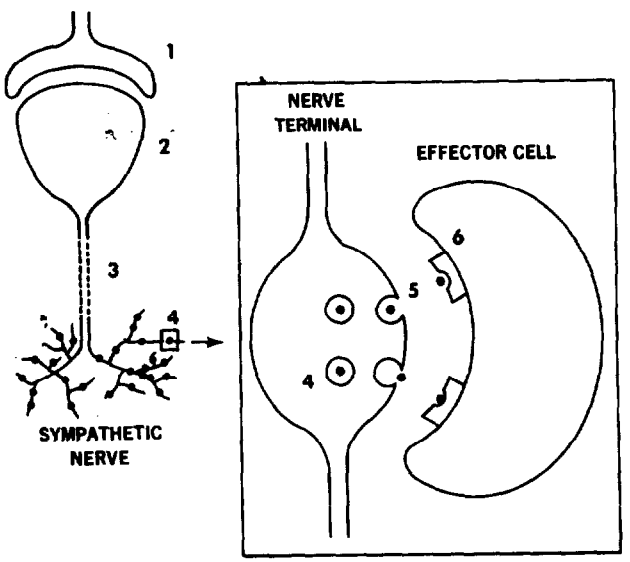

FIG. 2. Sympathetic nerves and the fate of noradrenaline. 1–6: Sites where the biosynthesis and action of the catecholamines are regulated. See text for explanation.

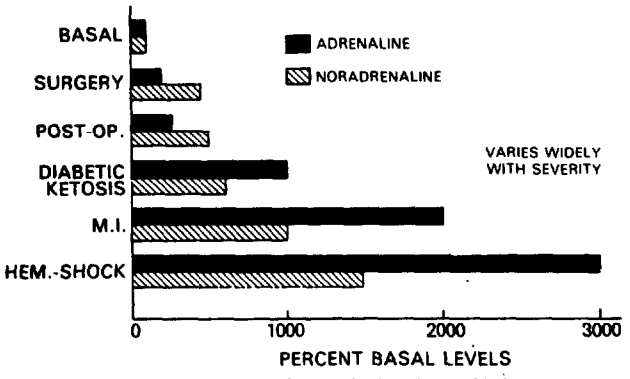

FIG. 3. Plasma levels of catecholamines during surgery and catastrophic illness. M.I. = myocardial infarction. (Data derived from several reviews.)

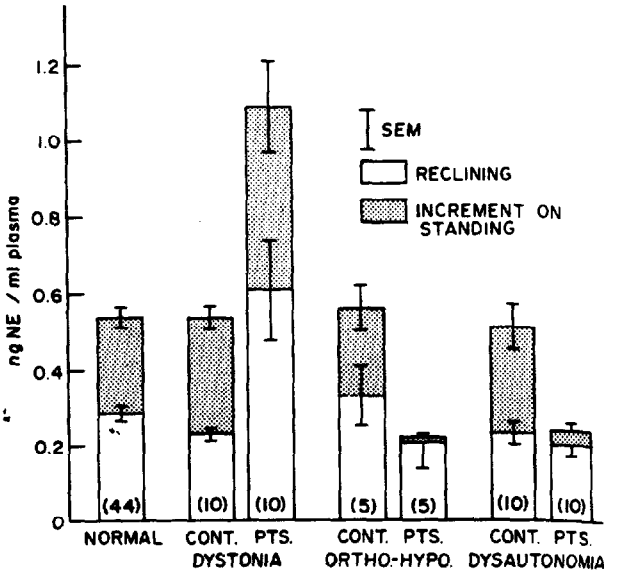

FIG. 4. Plasma levels of noradrenaline in patients (pts.) with autonomic disorders, compared with levels in controls (cont.) of the same age. (Data derived from several reviews.)

contents of the vesicle, including the catecholamine neurotransmitter and dopamine-β-hydroxylase, are discharged into the space between the varicosity and the effector organ. The transmitter then interacts with the adrenergic receptors on the cell surface of the effector organ. The noradrenaline liberated from the nerve terminal is inactivated mainly by reuptake into the nerve; some of the neurotransmitter is discharged into the blood stream and metabolized by catechol-O-methyltransferase and monoamine oxidase in the liver and kidney.

Because of the extremely low levels of noradrenaline and adrenaline in the blood, it has been difficult to accurately measure these amines until recently. Utilizing the enzyme catechol-O-methyltransferase and ^{3}H-methyl-S-adenosyl-methionine, it is now possible to measure noradrenaline and adrenaline in plasma. Noradrenaline reflects the activity of the sympathetic nervous system, and adrenaline that of the adrenal medulla. There is considerable elevation of noradrenaline and adrenaline after coffee drinking, smoking, public speaking, and stress. A massive discharge of noradrenaline and adrenaline is found after diabetic ketosis, myocardial infarction, hemorrhagic shock, and surgery[15] (Fig. 3). In dystonia patients, there is an elevation of plasma levels of catecholamine and reduced levels in dysautonomia. There is hardly any discharge of noradrenaline in orthostatic hypotension (Fig. 4).

Transmitters are in a dynamic state within the nerves. They are constantly in a state of flux. One can measure the turnover of the transmitter by injecting ^{3}H-noradrenaline and measuring the specific activity of the catecholamines in specific areas after various times. We noticed that in experimental hypertension there was a greater turnover of the catecholamine transmitters in the heart.[7] This was one of the earliest experiments showing the relationship of the sympathetic nervous system and hypertension. Most drugs used to treat hypertension affect the sympathetic nervous system.

Catecholamine Synthesis

The enzymes that synthesize catecholamines are found in the cell body nucleus and transported down the neuron to the varicosities by a process of axoplamic flow (Fig. 2). There are four enzymes involved in the synthesis of the catecholamines (Fig. 5). I would like to draw your attention to two. One is tyrosine hydroxylase and the other is phenylethanolamine N-methyltransferase (PNMT). Tyrosine hydroxylase converts the amino acid, tyrosine, to dopa in the nerves and is the rate-limiting enzyme for catecholamine synthesis. The other enzyme, PNMT, methylates noradrenaline to form adrenaline and is highly localized in the adrenal gland. Both of these enzymes are present in nerves and are highly regulated.

One important advance in the last few years is our increase in knowledge about the regulation of the synthesis of catecholamines. The adrenosympathetic system has the capacity to adapt to all kinds of normal and abnormal changes. Depending on the situation, tyrosine hydroxylase can rapidly adapt to synthesize more or less catecholamines. When nerve activity is reduced, the buildup of catecholamines in the nerves inhibits tyrosine hydroxylase by a classical negative feedback mechanism. If there is a great deal of nerve activity, there is less of the transmitter in the nerves and this enzyme is not restrained and more neurotransmitter is synthesized. We found that after prolonged nerve activity, such as stress, there is a gradual accumulation of tyrosine hydroxylase in the adrenal gland and in the sympathetic nerves. We wanted to find out what makes the level of tyrosine hydroxylase increase. Does the signal come from a presynaptic nerve or does it come from within the same nerve?

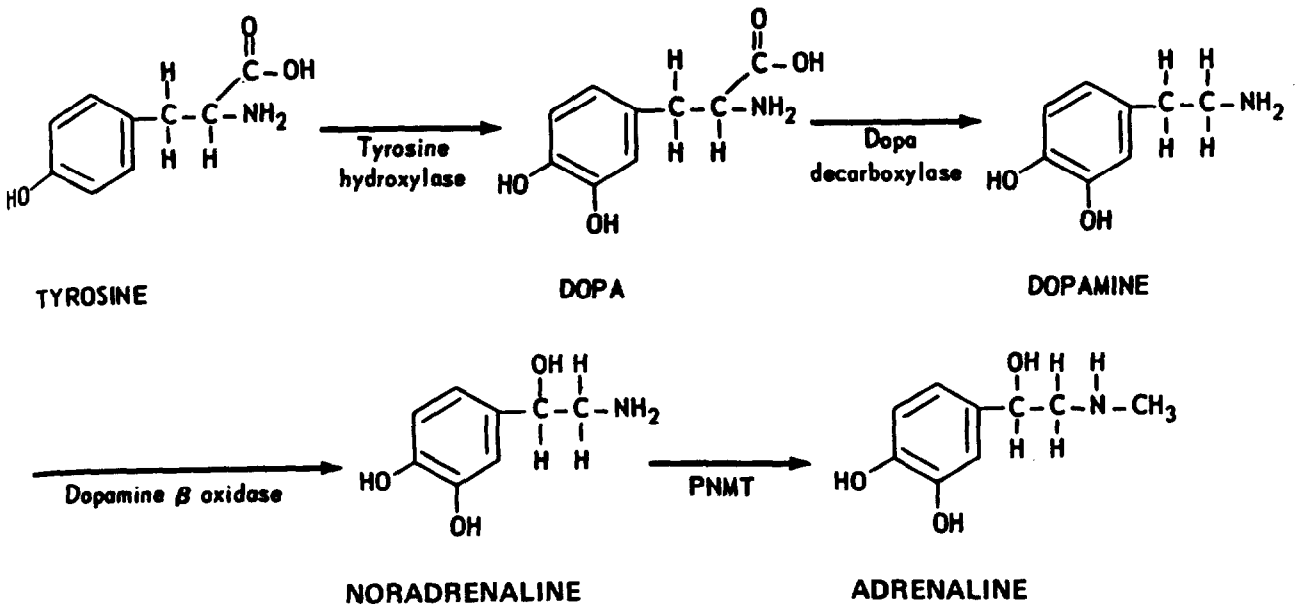

FIG. 5. Biosynthetic pathway of catecholamines.

We thought of a neat experiment to prove this. We decentralized the superior cervical ganglia of rats on one side and then subjected the animals to prolonged stress. After 24 hours, there was an increase in the tyrosine hydroxylase on the innervated side, but not on the denervated side. This indicated that the signal for increase in enzyme activity comes from presynaptic nerves (Fig. 2).

It has been recognized for many years that the adrenal cortex controls the synthesis of adrenaline in some unknown way. We had just characterized PNMT, the adrenaline-forming enzyme, and this provided an opportunity to examine how the adrenal cortex controls the synthesis of adrenaline. I think this next experiment would have been dear to the heart of Dr. Cushing. During an exchange of ideas with my then postdoctoral fellow, Richard Wurtman, we came up with a "doable" experiment. Wurtman, who is a better endocrinologist than I am, suggested that we remove the pituitary. Since the pituitary secretes adrenocorticotropic hormone (ACTH) and controls the cortical steroids in the adrenal cortex, we then could examine how hypophysectomy would affect PNMT in the adrenal medulla. When we hypophysectomized the rat, the adrenaline-forming enzyme in the adrenal gland went way down.[27] When either ACTH or dexamethasone was injected, the PNMT activity was restored. This clearly showed the interaction in adrenaline formation between these three organs: the pituitary, the adrenal cortex, and the adrenal medulla. In subsequent experiments we found that tyrosine hydroxylase and dopamine-β-hydroxylase in the adrenal medulla are mainly regulated by the splanchnic nerves. The adrenaline-forming enzymes are regulated by the pituitary and by the adrenal cortex.

Pineal Gland Studies

We learned a great deal about the regulation of effector organs by sympathetic nerves by studying the pineal gland. The pineal gland was recognized by the Greeks and they believed that it was the storage bin of thought because it was an unpaired structure in the brain. The philosopher Descartes elaborated on this concept. In his book on physiology "*De Homine*," Descartes proposed that the pineal gland played a central role in the action of the brain. In his formulation, the eye perceived an event of the real world and transmitted this information via strings in the brain to the pineal gland. This tilted the pineal gland and allowed animal humors to move down a hollow tube within the nerve to the muscle. When the humors reached the muscle, they caused it to swell and produce the appropriate response. With the hindsight of three centuries, it was a prophetic formulation of chemical neurotransmission. The pineal gland is jokingly referred to as the seat of the soul, and it was believed that its main purpose was to serve as a landmark for neurosurgeons and radiologists.

In the last 20 years or so, the pineal has been a very exciting organ to work on. About 50 years ago, two zoologists at Johns Hopkins found that an extract of the pineal gland caused tadpole skin to blanch. In 1958, Aaron Lerner, a dermatologist and biochemist, and his co-workers isolated the active blanching principal of the pineal gland and identified it as 5-methoxy N-acetylserotonin, and it was named "melatonin."

I read Lerner's paper in the *Journal of the American Chemical Society*[16] and was struck by several things about the chemical structure of melatonin. First, it had a methyl group attached to oxygen similar to that

of catecholamine metabolites. It also had a nucleus which resembled serotonin, another neurotransmitter. My colleagues and I worked out the biosynthesis of melatonin from tryptophan and serotonin, which I won't elaborate upon.[4] Another interesting facet of the pineal gland is that it undergoes circadian rhythms of serotonin and melatonin as well as the enzymes that synthesize melatonin. The pineal gland was found to be innervated by noradrenergic nerves and, of course, this also intrigued me. This led to many experiments showing how noradrenaline released from sympathetic nerves controls the synthesis of melatonin and circadian rhythms of indoleamines and its enzymes in the pineal.[4] I would like to describe one experiment to give the flavor of this type of work (Fig. 6). In collaboration with Solomon Snyder, a postdoctoral fellow, we devised experiments to find out whether sympathetic nerves control the pineal serotonin rhythm.[23] When we destroyed the sympathetic innervation of the pineal gland in rats by removal of the superior cervical ganglia, the serotonin rhythm was abolished. When the rats were kept in constant light, the serotonin rhythm disappeared. However, when they were in constant dark the rhythm persisted, indicating that there was an endogenous clock driving this rhythm. Later on, the work of Robert Moore showed that the driving oscillator for pineal rhythms, and many other rhythms, resides in the suprachiasmatic nucleus of the hypothalamus.

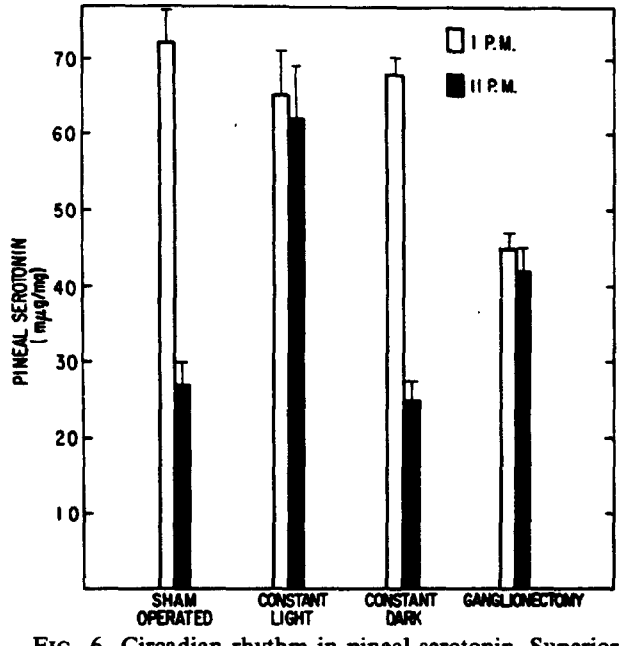

FIG. 6. Circadian rhythm in pineal serotonin. Superior cervical ganglia were removed bilaterally 7 days before rats were killed and the pineal glands examined for serotonin at the times and under the conditions indicated. (Composite of Fig. 1 from Snyder SH, *et al.*: Control of the circadian rhythm in serotonin content of the rat pineal gland. *Proc Natl Acad Sci USA 53*:301–305, 1965.)

We discovered that the rhythms in the pineal gland are due to diurnal changes in the release of noradrenaline from sympathetic nerves.[5] During the daytime, the release of noradrenaline is reduced. At night, there is an increased discharge of noradrenaline. With the reduced release of noradrenaline during the daytime, the β-adrenergic receptor of the pineal becomes more responsive. As night falls, there is an increased noradrenaline discharge onto a supersensitive receptor and an amplification of the response. There is a corresponding increase in the synthesis of serotonin N-acetyltransferase, the enzyme that makes melatonin.

Many problems in neurology and medicine involve changes in responsitivity of tissue receptors to neurotransmitters or hormones. These are becoming increasingly important. In other experiments, to show how the nerves control responsitivity of a cell, we used the rat pineal gland. This gland can be stimulated to make serotonin N-acetyltransferase by treatment with the β-adrenergic agonist, isoproterenol, in organ culture. When the pineal gland was denervated for 48 hours, removed, and treated with isoproterenol, it became 10 times more responsive in synthesizing the N-acetyltransferase. If we injected the rats with isoproterenol several times and then removed the gland and treated it with isoproterenol, the production of the melatonin-forming enzyme was much reduced. The receptor has a great capacity to adapt to too much or too little neurotransmitter (Fig. 7). With decreased nerve firing, there are more receptors and an increase of intracellular messengers, such as cyclic adenosine monophosphate (AMP). This cascade then amplifies a response with reduced neurotransmitter. When there is too much neurotransmitter, the reverse occurs and the response is dampened. This is an example of many years of research illustrating how one can get some very fundamental information and still have a good time during the work. The pineal gland is a wonderful organ to work with. Recently, methods have been developed that can measure very small amounts of melatonin in the blood. This makes it possible to do studies on melatonin, circadian rhythms, and β-adrenergic receptor responsitivity in man. It has already been found that man has the same type of rhythm in blood melatonin as was identified in the rat pineal, high at night and low during the daytime. Sudden exposure of humans to light at night causes a precipitous drop in the nighttime levels of plasma melatonin. The ability to measure melatonin in plasma now provides a new tool to study endocrine function and sympathetic nerve activity.

Research in the past 20 years has led us to conclude that the pineal gland is a neurochemical transductor.[4] Light signals from the eye are relayed along the retinohypothalamic tract to the suprachiasmatic nucleus. The suprachiasmatic nucleus is believed to be the circadian oscillator. The suprachiasmatic nucleus

sends nerve impulses to the superior cervical ganglion, which in turn stimulates noradrenergic nerves to release noradrenaline. Thus, noradrenaline activates β-adrenergic receptors on the pineal cell surface. The interaction of noradrenaline with the β-adrenergic receptor transmits the neurotransmitter message through the membrane to stimulate the production of the enzyme which makes melatonin. I am sure that the elucidation of the noradrenaline message to tell the pineal gland to secrete melatonin can give us insight about the action of other nerves.

Psychoactive Drugs

It has been known now for the last 15 years or so, that the brain has specific tracts that contain noradrenaline and dopamine and that they are involved in normal function as well as diseases, such as Parkinson's, Huntington's, and possibly many other neurological and endocrinological abnormalities. Our knowledge about neurotransmitters and their function in the brain is just beginning. We were very much interested in knowing whether the fundamental work we did on the peripheral noradrenergic nervous system would also apply to the central nervous system, particularly with respect to drugs that affect behavior.

One group of drugs which we were very much interested in was the antidepressant drugs. We found that tricyclic antidepressant drugs that are clinically effective also blocked the reuptake of noradrenaline in brain tissue[10] (Fig. 8). As I described in the early part of my talk, reuptake of noradrenaline inactivated this neurotransmitter. Compounds that were clinically ineffective but had related chemical structures did not block uptake. Monoamine oxidase inhibitors block the metabolic breakdown of the catecholamines and increase the level of these amines in the brain. Monoamine oxidase inhibitors are also effective in the treatment of some depressions. Thus, drugs that decrease catecholamines cause depression, whereas drugs that elevate these amines relieve depression. These observations generated a lot of research among biological psychiatrists and led to the proposal of the catecholamine hypothesis of affective disorders.[21] Although the hypothesis is somewhat simplistic, it provided a useful framework whenever new approaches to the understanding of depression were sought.

Another important development in psychiatry was the demonstration of the relationship between psychoses and dopamine. It has been shown that dopamine has its own receptor in the brain and that antipsychotic drugs block this receptor.[14] Experiments by Snyder and co-workers[6] showed that the order of potency of antipsychotic drugs competing for the dopamine receptor is in the same order as their average clinical dosage, again implicating dopamine in schizophrenic disorders. This sort of experiment would have been unthinkable 20 or 30 years ago.

In the course of our work in the last few years, we found tracts in the brain containing adrenaline, mainly in the brain stem.[20] How we learned this was to punch out very small areas of the brain by a technique devised by my colleague, M. Palkovits. Brain punches from the A_1 and A_2 areas of the brain stem contained PNMT, the enzyme that makes adrenaline. Specific adrenergic tracts in the A_1 and A_2 areas of the brain stem could be visualized by a technique using a fluorescent antibody to the adrenaline-forming enzyme.[13] It appears that these adrenergic tracts are involved in the regulation of blood pressure. There was an increase in the adrenaline-forming enzyme in the A_1 and A_2 regions of the brain stem in experimental hypertension produced by deoxycorticosterone acetate (DOCA) and salt and also in spontaneous hypertension.[19] When a compound that blocks PNMT was given to hypertensive rats, a fall in blood pressure resulted. These experiments tell us a great deal about the role of the adrenaline-containing tract in control-

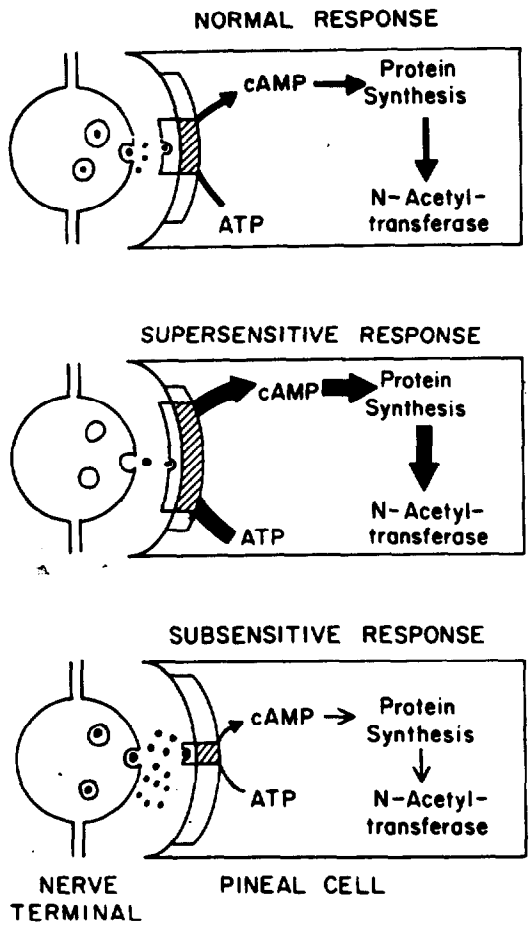

FIG. 7. Neural control of β-adrenergic receptor and the synthesis of the melatonin-forming enzyme N-acetyltransferase. cAMP = cyclic adenosine monophosphate; ATP = adenosine triphosphate. (Reproduced with permission from Axelrod J: The pineal gland: a neurochemical transducer. *Science 184:*1341–1348, 1974.)

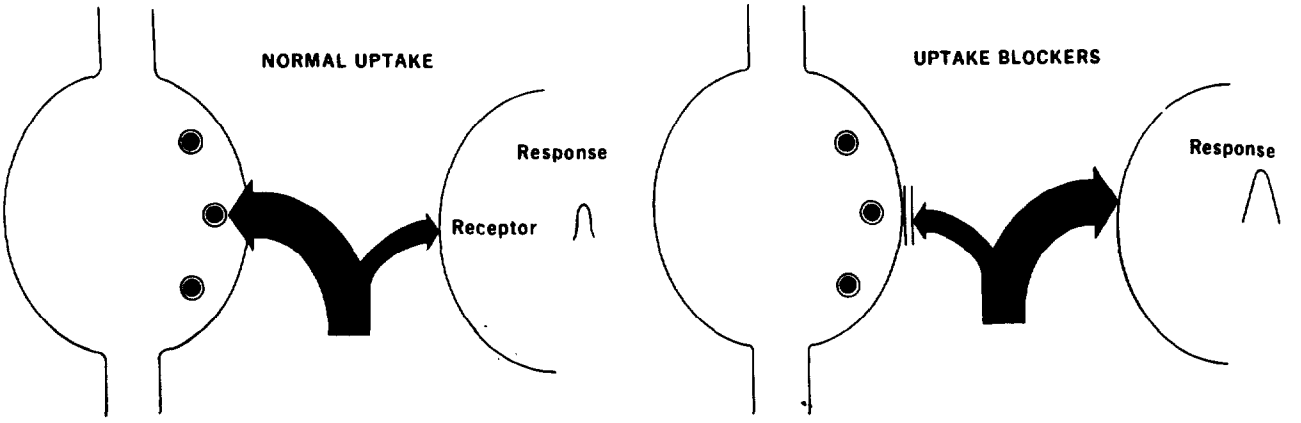

FIG. 8. Effect of antidepressant drugs on the uptake of noradrenaline in the noradrenergic nerve terminal.

ling blood pressure, and perhaps something about new therapeutic agents to treat hypertension.

The field of neurotransmitters has exploded enormously in the last few years with the recognition of many neuropeptides and amino acids having the properties of neurotransmitters.[22] These recent discoveries indicate the complexity with which the brain functions. It now appears that the brain can function as an enormous endocrine organ. Nerves can release numerous endocrine-like substances, encephalins, thyroid-releasing hormone, neurotension, adrenocorticotropic hormone, etc., which have neurotransmitter-like properties. There is a great deal to be done in neurotransmitter research, and there are many good neuroscientists to do the work.

References

1. Armstrong MD, McMillan A: Identification of a major urinary metabolite of norepinephrine. **Fed Proc 16:**146, 1957 (Abstract)
2. Axelrod J: Noradrenaline: fate and control of its biosynthesis. **Science 173:**598–606, 1971
3. Axelrod J: O-methylation of epinephrine and other catechols in vitro and in vivo. **Science 126:**400–401, 1957
4. Axelrod J: The pineal gland: a neurochemical transducer. **Science 184:**1341–1348, 1974
5. Brownstein M, Axelrod J: Pineal gland: 24-hour rhythm in norepinephrine turnover. **Science 184:**163–165, 1974
6. Creese I, Burt DR, Snyder SH: Dopamine receptor binding predicts clinical and pharmacological potencies of antischizophrenic drugs. **Science 192:**481–483, 1976.
7. de Champlain J, Krakoff LR, Axelrod J: Catecholamine metabolism in experimental hypertension in the rat. **Circ Res 20:**136–145, 1967
8. Elliot TR: On the action of adrenaline. **J Physiol (Lond) 31:**xx–xxi, 1904 (Proceedings)
9. Falck B, Hillarp N-Å, Thieme G, et al: Fluorescence of catechol amines and related compounds condensed with formaldehyde. **J Histochem Cytochem 10:**348–354, 1962
10. Glowinski J, Axelrod J: Inhibition of uptake of tritiated-noradrenaline in the intact rat brain by imipramine and structurally related compounds. **Nature 204:**1318–1319, 1964
11. Hertting G, Axelrod J, Kopin IJ, et al: Lack of uptake of catecholamines after chronic denervation of sympathetic nerves. **Nature 189:**66, 1961
12. Hoffer A, Osmond H, Smythies J: Schizophrenia: a new approach; result of year's research. **J Ment Sci 100:** 29–45, 1954
13. Hökfelt T, Fuxe K, Goldstein M, et al: Immunohistochemical evidence for the existence of adrenaline neurons in the rat brain. **Brain Res 66:**235–251, 1974
14. Kebabian JW, Petzold GL, Greengard P: Dopamine-sensitive adenylate cyclase in caudate nucleus of rat brain, and its similarity to the "dopamine receptor." **Proc Natl Acad Sci USA 69:**2145–2149, 1972
15. Kopin IJ: Plasma levels of catecholamines and dopamine β-hydroxylase, in Trendelenburg U, Weiner N (eds): **Catecholamines II.** Berlin: Springer-Verlag, 1981 (In press)
16. Lerner AB, Case JD, Takahashi Y, et al: Isolation of melatonin, the pineal gland factor that lightens melanocytes. **J Am Chem Soc 80:**2587, 1958 (Abstract)
17. Loewi O: Über humorale Übertragbarkeit der Herznervenwirkung. **Pfluegers Arch 189:**239–242, 1921
18. Rosell S, Kopin IJ, Axelrod J: Fate of H^3-noradrenaline in skeletal muscle before and following sympathetic stimulation. **Am J Physiol 205:**317–321, 1963
19. Saavedra JM, Grobecker H, Axelrod J: Adrenaline-forming enzyme in brainstem: elevation in genetic and experimental hypertension. **Science 191:**483–484, 1976
20. Saavedra JM, Palkovits M, Brownstein MJ, et al: Localisation of phenylethanolamine N-methyl transferase in the rat brain nuclei. **Nature 248:**695–696, 1974
21. Schildkraut JJ: The catecholamine hypothesis of affective disorders: a review of supporting evidence. **Am J Psychiatry 122:**509–522, 1965
22. Snyder SH: Brain peptides as neurotransmitters. **Science 209:**976–983, 1980
23. Snyder SH, Zweig M, Axelrod J, et al: Control of the circadian rhythm in serotonin content of the rat pineal gland. **Proc Natl Acad Sci USA 53:**301–305, 1965
24. von Euler US: A specific sympathomimetic ergone in adrenergic nerve fibres (sympathin) and its relations to adrenaline and nor-adrenaline. **Acta Physiol Scand 12:** 73–97, 1946
25. Whitby LG. Axelrod J, Weil-Malherbe H: The fate of

H^3-norepinephrine in animals. **J Pharmacol Exp Ther 132:**193–201, 1961

26. Wolfe DE, Potter LT, Richardson KC, et al: Localizing tritiated norepinephrine in sympathetic axons by electron microscopic autoradiography. **Science 138:** 440–442, 1962
27. Wurtman RJ, Axelrod J: Control of enzymatic synthesis of adrenaline in the adrenal medulla by adrenal cortical steroids. **J Biol Chem 241:**2301–2305, 1966

Manuscript received July 27, 1981.

The 1981 Harvey Cushing ovation was delivered at the Annual Meeting of the American Association of Neurological Surgeons, Boston, Massachusetts, on April 7, 1981.

Address reprint requests to: Julius Axelrod, Ph.D., Chief, Pharmacology Section, Laboratory of Clinical Science, National Institute of Mental Health, Bethesda, Maryland 20014.